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Transferosomes: A Promising Vesicular Carrier in Enhancing Drug Permeability

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Abstract

Transferosomes are specialized lipid-based vesicles designed to improve drug delivery through the skin. Their unique flexibility allows them to penetrate the skin's protective barrier more efficiently than traditional transdermal systems. Transferosomes combine the characteristics of liposomes and niosomes because they contain liposomes (phospholipids and cholesterols) and niosomes as components (non-ionic surfactants; edge activators). As a result, they are referred to as the first generation of elastic liposomes. However transdermal drug delivery is difficult due to the presence of the skin's protective barrier, transferosomal drug delivery overcomes all obstacles due to its unique characteristics, such as its ultra-deformable vesicular nature. Transdermal drug delivery systems (TDDS), which are self-administrable and non-invasive, can improve bioavailability and patient compliance by bypassing first-pass metabolism. Vesicular-based TDDS have attracted a lot of attention in recent years because they're designed for controlled, efficient, and targeted drug delivery. One of these delivery technologies, transpersonal-based formulations, has grown in popularity due to its ability to achieve all of the desired criteria and quality.

Keywords: Transferosomes; Transdermal; Phospholipids; Penetration; Ultra-Deformable

Abbreviations

TDDS: Transdermal Drug Delivery Systems; CSLM: Confocal Scanning Laser Microscopy; PCS: Photon Correlation Spectroscopy; DLS: Dynamic Light Scattering; TEM: Transmission Electron Microscopy; EE: Entrapment Efficiency; HPLC: High-Performance Liquid Chromatography; API: Active Pharmaceutical Ingredient; ICH: International Conference on Harmonization; RH: Relative Humidity.

Introduction

Today's research environment is on creating novel drug delivery systems that have a prominent level of therapeutic efficacy and are well-accepted by patients. When placed to the intended spot, a transferosome is a tool or equipment that facilitates the simple penetration of the skin to transfer medications [1]. The preferred form of transferosomes is an ultra-deformable vesicle with an aqueous core surrounded

by a complex lipid bilayer. Even in cases where the pores are far smaller than the vesicles' size, transferosome vesicles may yet effectively traverse microporous barriers [2].

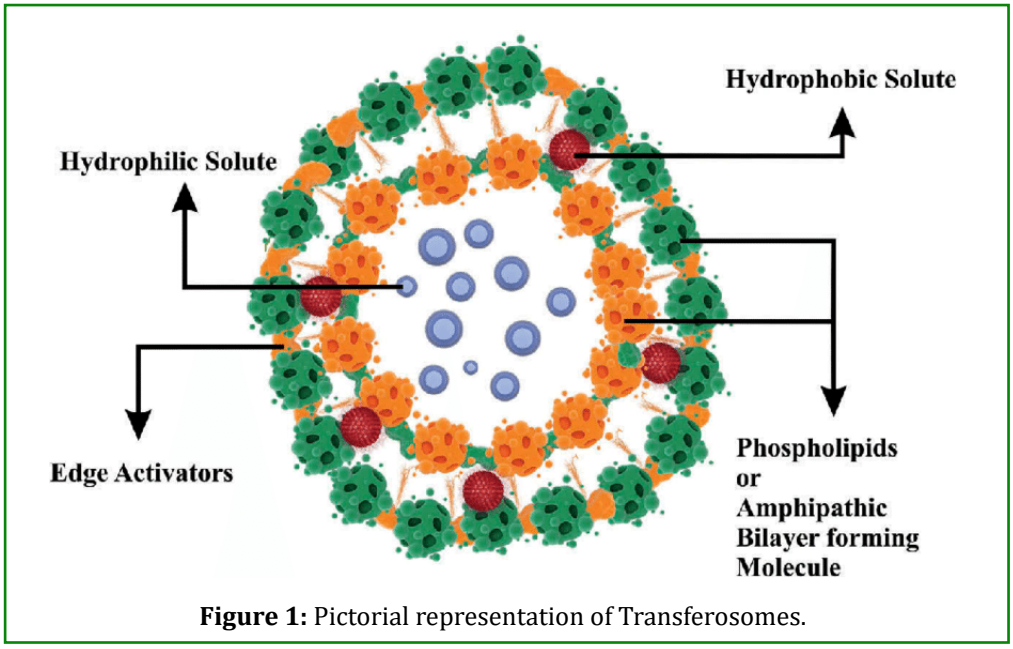

Figure 1: Pictorial representation of Transferosomes.

Targeted drug delivery is made possible by transferosomes' ability to effectively cross a variety of transport barriers and function as an effective drug carrier for the prolonged release of therapeutic medicines [3]. Because "edge activators," which include surfactants like sodium cholate, sodium deoxycholate, span 80, and tween 80, have been added, the lipid bilayer surrounding the inner layer of transferosomes has been particularly changed." Transferosomes are a new and innovative drug delivery system that was developed to solve the problem of clarification. They are specific types of liposomes that contain or have phosphatidylcholine water, an edge activator, and they can penetrate the skin barrier along the transcutaneous gradient. Due to its ultra-flexible and self-optimized membrane, transferosomes are mostly used in skin care [4].

Materials and Methods

Materials Required for Preparation of the Transferosomes

Phospholipids: (Vesicles forming component) Ex. - Soya phosphatidylcholine, egg phosphatidylcholine.
Surfactant: (Providing flexibility). Ex. - Sod.deoxycholate, Tween-80, Span-80.
Alcohol: (As a solvent). Ex. - Ethanol, methanol.
Buffering Agent: (As a hydrating medium) Ex. - Saline phosphate buffer (pH 6.4).
Dye: {for Confocal scanning laser microscopy (CSLM)} Ex. – Rhodamine.

Method of Preparation of Transfersomes

There is no standard preparation protocol or set formula for this process, even though there are several patented transfersome preparation techniques. Therefore, to obtain the most appropriate carriers with the best deformability, drug-carrying capacity, and stability, the optimal preparation conditions and vesicle compositions must be identified, designed, and optimized by conducting individually designed experimental procedures for each therapeutic agent [5]. The thin film hydration approach, sometimes referred to as the rotary evaporation-sonication method, is the traditional way of preparing transfersomes. Vortexing-sonication, the modified handshaking procedure, centrifugation, suspension homogenization, reverse-phase evaporation, high-pressure homogenization, and ethanol injection are further modified preparation techniques. The following is a general description of each method:

Thin Film Hydration Technique: In a round-bottom flask, the phospholipids and edge activator (the components that form vesicles) are dissolved using a combination of volatile organic solvents (for instance, methanol and chloroform in an appropriate (v/v) ratio). This stage might include the incorporation of the lipophilic medication. A rotating vacuum evaporator is used to evaporate the organic solvent above the lipid transition temperature under decreased pressure to generate a thin layer. Sustain it under Hoover to get rid of any remaining solvent residue. After the thin film has been formed, it is hydrated by rotating it for the necessary amount of time at a suitable temperature using a

buffer solution with the proper pH (for instance, pH 7.4). At this point, the hydrophilic drug inclusion can be completed. To create tiny vesicles, the resultant vesicles are sonicated in a bath or probe sonicator after being swelled at ambient temperature. Extrusion across a sandwich of 200 nm to 100 nm polycarbonate membranes homogenizes the sonicated vesicles [2,6].

Modified Hand Shaking (Lipid Film Hydration Technique): The method comprises of the following steps: - The rotating evaporation-sonication method and the modified handshaking method have the same fundamental idea. The organic solvent, lipophilic medication, phospholipids, and edge activator are added to a round-bottom flask during the modified handshaking procedure. The solvent should entirely dissolve each excipient, yielding a transparent, clear solution. Then, rather than employing a rotating vacuum evaporator, the organic solvent is eliminated by evaporation while shaking hands. The round-bottom flask is kept partially submerged in a water bath that is kept at a high temperature (for instance, 40–60 °C) in the interim. Within the flask wall, a thin lipid coating subsequently forms. The solvent is allowed to completely evaporate in the flask overnight. After that, the produced film is gently shaken and hydrated with the suitable buffer solution at a temperature higher than its phase transition temperature. At this point, the hydrophilic drug inclusion can be completed [6].

Vortexing-Sonication Method: In a phosphate buffer, the medication, edge activator, and phospholipids are combined. After this, the mixture is vortexed to produce a milky transfersomal suspension. After being sonicated for the appropriate amount of time at room temperature using a bath sonicator, it is extruded through polycarbonate membranes (450 and 220 nm, for example) [7].

Suspension Homogenization Method: To make transfersomes, combine a suitable quantity of edge activator with an ethanolic phospholipid solution. After the suspension is created, it is combined with buffer to obtain the total lipid concentration. After that, the mixture is frozen, thawed, and sonicated two to three times, respectively [8].

Reverse-Phase Evaporation Method: In a round-bottom flask, the phospholipids and edge activator are combined with an organic solvent mixture (diethyl ether and chloroform, for example) and dissolved. This stage can include the incorporation of the lipophilic medication. The lipid films are then obtained by utilizing a rotary evaporator to evaporate the solvent. The organic phase, which primarily consists of isopropyl ether and/or diethyl ether, is where the lipid films are re-dissolved. The organic phase is then combined with the aqueous phase to create a two-phase system. At this point, the hydrophilic drug inclusion can be completed. After that, this system is sonicated with a bath sonicator until a uniform water-based emulsion forms. Using a rotary evaporator, the organic solvent is gradually evaporated to create a viscous gel that eventually turns into a vesicular suspension [9].

High-Pressure Homogenization Technique: In PBS or distilled water with alcohol, the phospholipids, edge activator, and medication are evenly distributed. They are then concurrently agitated and subjected to ultrasonic shaking. After that, the mixture is shaken intermittently with ultrasonic technology. A high-pressure homogenizer is then used to homogenize the resultant mixture. Lastly, the transfersomes are kept in suitable storage settings [10].

Ethanol Injection Method: The phospholipid, edge activator, and lipophilic medication are dissolved in ethanol with magnetic stirring for the appropriate amount of time to yield a transparent solution, which creates the organic phase. The water-soluble materials are dissolved in the phosphate buffer to create the aqueous phase. At this point, the hydrophilic drug inclusion can be completed. The temperatures of both solutions reach 45–50 °C. Subsequently, the aqueous solution is continuously stirred for the designated amount of time while the ethanolic phospholipid solution is added dropwise. The process of removing ethanol involves placing the resulting dispersion into a vacuum evaporator and sonicating to reduce the particle size [11].

Mechanism of Penetration of Transferosomes

Transferosomes, when used properly, may transfer 0.1 mg of lipid per hour and square centimetre area across undamaged skin. This number is significantly higher than what is generally produced via transdermal concentration gradients. The reason for this high flow rate is due to naturally occurring "transdermal osmotic gradients," which means that another far more significant gradient exists across the skin. The skin penetration barrier creates an osmotic gradient that keeps the viable portion of the epidermis (75% water content) and the almost dry stratum corneum (15%) close to the skin's surface from losing water [12]. This gradient also keeps the skin from drying out. Even at unphysiologically high transdermal water loss, ambient air acts as a perfect sink for the water molecule, maintaining the gradient's stability. Every polar lipid draws some water to it. This is because the hydrophilic lipid residues and their proximal water interact in an energetically beneficial way. The majority of lipid bilayers therefore naturally withstand induced dehydration. As a result, all polar lipid vesicles-derived lipid vesicles migrate from relatively dry locations to regions with a high enough concentration of water. Lipid vesicles sense this "osmotic gradient" and attempt to avoid total drying by traveling along it when lipid suspension (transferosome) is applied to skin that has partially dried due to water evaporation loss. Less deformable vesicles, such as standard liposomes, are confined to the skin's surface where they completely dehydrate and fuse, giving them less penetration power than transferosomes. Only sufficiently deformable vesicles can pass through the skin's narrow pores because surfactant-based transferosomes have more suitable rheological and

hydration properties than that which accounts for their greater deformability. In this way, transferosomes are maximized in their flexibility, allowing them to fully exploit the trans epidermal osmotic gradient (water concentration gradient). Transferosomes squeeze themselves along the intracellular sealing lipids of the stratum corneum to get around the barrier of skin penetration [13].

The current state of knowledge regarding how to improve the delivery of active substances through and into the skin is lacking. Two different action mechanisms have been suggested [14].

- At the skin's surface, transferosomes function as drug vectors and stay whole.
- Transferosomes serve as penetration enhancers, breaking up the stratum corneum's highly structured intercellular lipids to allow drug molecules to enter and pass through the layer more easily.

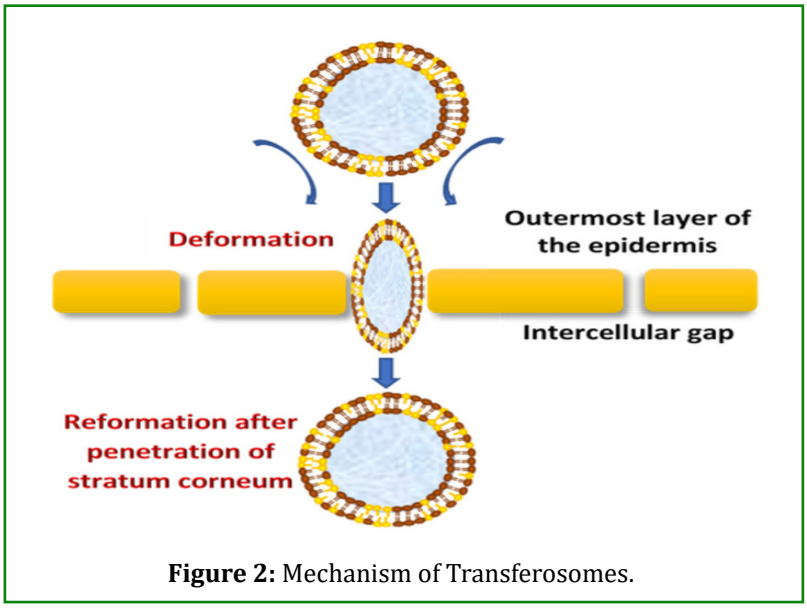

Figure 2: Mechanism of Transferosomes.

The initial mechanism was postulated by Cevc and colleagues. They proposed that deformable liposomes traverse the epidermis and enter the systemic circulation after penetrating the stratum corneum due to the transdermal moisture gradient that is generally present in the skin. According to current studies, the two mechanisms work together to cause the vesicles to penetrate and permeate the skin. One of the two methods predominates, depending on the composition of the transferosomes and the type of the active material (lipophilic or hydrophilic) [15].

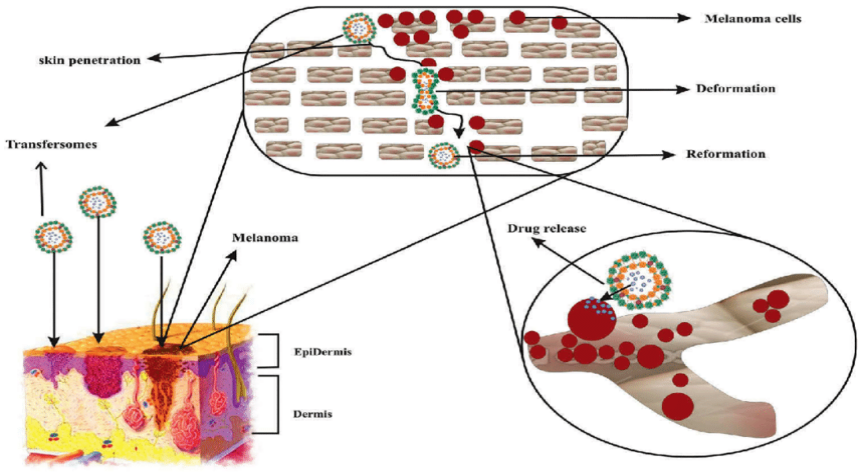

Figure 3: Diagrammatic representation of transferosome's mode of action for transdermal anticancer drug delivery.

Characterization of the Transferosomes

The vesicle shape and size, size distribution, polydispersity index, zeta potential, number of vesicles for cubic mm, entrapment efficiency, degree of deformability, and skin permeability measurements are among the published methods used to determine the characterization parameters of the transfersomes. These methods are helpful for optimizing the transfersomal formulation. A detailed explanation of each characterisation technique is provided below [16].

Vesicle Size, Zeta Potential and Morphology

One of the key variables in the creation of transfersomes, batch-to-batch comparisons, and scale-up procedures is the vesicle size. The vesicle size fluctuation during storage is a significant factor affecting the formulation's physical stability. Because of their high curvature state bilayer membranes, vesicles smaller than 40 nm are more likely to undergo fusion processes, while considerably bigger and electro neutral transfersomes aggregate by van der Waals interactions because of their comparatively larger membrane contact surfaces. One element influencing the drug compounds' ability to be encapsulated in transfersomes is the size of the vesicles. A greater aqueous core volume is preferable for the encapsulation of hydrophilic chemicals, whereas a higher lipid-to-core ratio is chosen for lipophilic and amphiphilic agents. The vesicle diameter can generally be found using photon correlation spectroscopy (PCS) or the dynamic light scattering (DLS) approach. It is possible to combine the vesicle suspension with a suitable medium and measure the vesicular size in triplicate. Additionally, the sample can also be made in distilled water and filtered using a 0.2 mm membrane filter as an alternative method. In order to measure the size of the vesicles using DLS or PCS, the filtered sample is diluted with filtered saline. Furthermore, although transmission electron microscopy (TEM) is employed to examine structural changes, the DLS method-associated computerized inspection system by Malvern Zetasizer can be used to determine the vesicle size and size distribution. Malvern Zetasizer is used in the electrophoretic mobility technique to detect the zeta potential. Phase contrast microscopy or transmission electron microscopy (TEM) can be used to visualize transfersome vesicles [17].

Number of Vesicles per Cubic mm

The optimization of the transfersome composition and other process variables depends on this parameter. Five times improperly transferred somal compositions are diluted with 0.9% sodium chloride. This sample is examined using an optical microscope and a haemocytometer. Under an optical microscope, the transfersomes with vesicles larger than 100 nm can be seen [18,19].

Entrapment Efficiency (%EE)

The quantity of drug entrapped in the formulation is known as the entrapment efficiency (%EE). By removing the unentrapped medication from the vesicles using a variety of methods, including mini-column centrifugation, the EE is ascertained. The %EE in this procedure can be ascertained using direct or indirect approaches. The direct method would be to remove the supernatant from ultracentrifugation and then use a suitable solvent that can lyse the sediment to rupture the sedimented vesicles. The contaminants can then be eliminated by diluting and filtering the resultant solution with a syringe filter (0.22–0.45 μm). Using analytical techniques, such as spectrophotometry or modified high-performance liquid chromatography (HPLC), the drug content is ascertained based on the analytical technique used for the active pharmaceutical ingredient (API) [20].

Degree of Deformability

This parameter is significant since it influences the transfersomal formulation's penetration. Pure water is used as the benchmark in this investigation. The mixture is run through many microporous filters with known pore diameters ranging from 50 to 400 nm. Using DLS measurements, the particle size and size distribution are recorded following each pass [3,18].

In Vitro Drug Release

A scientific method to optimize the transfersomal formulation can be made possible by the in vitro drug release profile, which can offer essential information on the formulation design as well as specifics on the release mechanism and kinetics. Transfersomes' in vitro drug release is usually assessed in relation to the free drug or the reference product. Clearly, a number of investigations have produced fruitful information about the drug release characteristics of created transfersome formulations [21].

Stability of Transfersomes

The structure and size of the vesicles in relation to time can be used to determine the stability of transfersome vesicles. The mean size and structural changes can be found using DLS and TEM, respectively. The tailored transfersomal formulations can be kept at various temperatures in hermetically sealed amber vials. According to ICH (International Conference on Harmonization) guidelines, In the context of stability testing novel medicinal substances and products, the standard storage conditions are defined as follows: 25 ± 2 °C/60% relative humidity (RH) ± 5% RH for a 12-month period, or 30 ± 2 °C/65% RH ± 5% for an expedited testing period of 40 ± 2 °C/75% RH ± 5% for six months. It is recommended that drug items meant for refrigeration undergo long-term

storage at 5 ± 3 °C for a duration of 12 months, followed by an expedited study at 25 ± 2 °C/60% RH ± 5% RH for a period of six months. Failure to meet the drug product's standards is considered a substantial change.

Advantages of Transferosomes

- Transferosomes have an infrastructure made up of both hydrophobic and hydrophilic moieties, they can hold medicinal molecules with a broad variety of solubility. They do not suffer appreciable loss when they bend and pass through thin constriction (between 5 and 10 times smaller than their own diameter) [22,23].
- This system's high deformability allows intact vesicles to penetrate more effectively. Both low and large molecular weight medications, such as insulin, albumin, sex hormone, corticosteroids, analgesics, and anaesthetics, can be transported by them.
- They are similar to liposomes in that they are made of natural phospholipids, making them both biocompatible and biodegradable.
- When it comes to lipophilic drugs, their entrapment efficiency is close to 90%.
- They shield the medication within the capsule from being broken down by proteins and peptides, for example.
- They serve as depots, releasing their contents gradually and being able to be employed for topical and systemic medication administration. Because of their straightforward process and lack of needless or inappropriately pharmacological ingredients, they are simple to scale up.
- Transferosomes exhibit negligible loss during deformation and can squeeze through constrictions that are five to ten times smaller than their own diameter. Better penetration of intact vesicles across tight junctions is facilitated by this high deformability.
- Disadvantages of Transferosomes [22,23].
- Transferosomes are chemically unstable because of their predisposition to oxidative degradation.
- Purity of natural phospholipids is another criterion militating against adoption of transferosomes as drug delivery vehicles.
- Transferosomes formulations are expensive.

Applications of Transferosomes

Applications of transferosomes in transdermal medication delivery have been thoroughly researched during the past few decades. The section below provides descriptions of a few of these uses.

Delivery of Antioxidants: Using a modified thin-film hydration method and high-pressure homogenization technique, Avadhani, et al. [24] created nanotransfersomes containing EGCG and hyaluronic acid to improve their efficacies as UV radiation protectors, antioxidants, and antiaging substances [24]. Wu, et al. [10] created transferosomes with resveratrol in 2019 by employing the high-pressure homogenization method. It was discovered that the transferosomes that were produced could increase resveratrol's stability, bioavailability, solubility, and safety.

Delivery of Anticancer Drugs: The topical treatment of melanoma using transferosome embedded oligopeptide hydrogels containing paclitaxel made using the thin-film dispersion approach was the subject of a 2018 study by Jiang, et al. [25] Tumor tissues were demonstrated to be efficiently penetrated by transferosomes consisting of phosphatidylcholine, tween80, and sodium deoxycholate [25].

Delivery of Corticosteroids: Cevc and Blume conducted research in 2003 and 2004 on the biological activity and properties of halogenated corticosteroid triamcinolone-acetonide-loaded transferosomes made using the traditional thin-film hydration method. The outcomes demonstrated that transferosomes had a lower therapeutic dosage, longer duration of action, and enhanced biological potency [26].

Delivery of Anti-Inflammatory Drugs: Several research groups created and investigated diclofenac sodium, celecoxib, mefenamic acid, and curcumin-loaded transferosomes for topical delivery. According to research findings, transferees may enhance the anti-inflammatory medications' stability and effectiveness [27].

Conclusion

Transferosomes offer a significant advancement in transdermal drug delivery systems by enhancing the permeability and efficacy of drug delivery through the skin. Their flexible, deformable nature allows for better penetration into deeper skin layers, leading to improved therapeutic outcomes and controlled drug release. Despite their potential benefits, challenges such as production scalability and quality consistency need to be addressed. Overall, transferosomes hold promise for revolutionizing transdermal drug delivery, making treatments more effective and minimizing side effects.

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